# Metabolism of Sulfometuron Methyl in Lactating Goats

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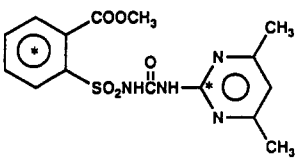
Sulfometuron methyl, the active ingredient in Oust herbicide, was rapidly metabolized and excreted in two goats. After 7 days of dosing two goats with [$^{14}$C]sulfometuron methyl at a dietary level of 25 or 60 ppm, greater than 93% of the dose was excreted in the urine and feces. Approximately 60% of the dose rapidly metabolized to (hydroxymethyl)pyrimidine sulfometuron methyl and was excreted in the urine. Cleavage of the sulfonylurea bridge did occur, producing primarily sulfonamide and (hydroxymethyl)pyrimidinamine. Accumulation of $^{14}$C-labeled residues in milk, meat, and fat was minimal. Only 24–44% of the $^{14}$C-labeled residues in the liver were extractable using aqueous or organic solvents. An additional 50–70% of the $^{14}$C-labeled residues were solubilized by proteolytic enzyme, indicating the association of these $^{14}$C-labeled residues with protein. The kidney had similar percentages of protein-bound $^{14}$C-labeled residues.

## INTRODUCTION

The sulfonylurea herbicide sulfometuron methyl [methyl 2-[[[[(4,6-dimethyl-2-pyrimidinyl)amino]carbonyl]-amino]sulfonyl]benzoate] is the active ingredient of Oust herbicide. An effective weed-control agent, Oust is




sulfometuron methyl

(*denotes location of $^{14}$C)

registered for the control of unwanted vegetation on rights-of-way and industrial sites and for other noncrop usage. A few ounces per acre control over 60 weed species.

Because of the general use of Oust Herbicide, it is important to understand its metabolic pathway in animals, especially ruminants. This study describes the metabolic fate of sulfometuron methyl in the goat, a representative ruminant animal. Previous publications (Anderson and Dulka, 1985; Harvey et al., 1985) discussed some properties of sulfometuron methyl that could affect the herbicide's environmental and metabolic fate in soil.

## MATERIALS AND METHODS

[*pyrimidine*-2-$^{14}$C]- and [*phenyl*(U)-$^{14}$C]sulfometuron methyl were synthesized at New England Nuclear (Boston, MA) and at Du Pont Agricultural Products (Wilmington, DE), respectively. The [*pyrimidine*-2-$^{14}$C]sulfometuron methyl and [*phenyl*(U)-$^{14}$C]sulfometuron methyl had specific activities of 30.5 and 12.0 μCi/mg, respectively, and radiochemical purities of 99%. All metabolite reference standards were synthesized at Du Pont Agricultural Products. Unless specified otherwise, all solvents used were of HPLC grade; all other solvents and common chemicals were of reagent grade or better. All solvents and chemicals were handled, stored, and disposed of according to recommended procedures listed in the Material Safety Data Sheet from each manufacturer.

**Preparation of Capsules.** A solution of [*pyrimidine*-2-$^{14}$C]-sulfometuron methyl was prepared by dissolving 26.5 mg of [*pyrimidine*-2-$^{14}$C]sulfometuron methyl and 374.2 mg of non-radiolabeled sulfometuron methyl in 32 mL of dichloromethane. The specific activity was verified by liquid scintillation counting (LSC). Two milliliters of this solution was pipetted into each of 15 gelatin capsules (size 00, Eli Lilly, Indianapolis, IN), and the dichloromethane was removed under a stream of nitrogen. After the dichloromethane had evaporated, each capsule was capped and embedded in a larger gelatin capsule containing 6 g of goat chow. The total [*pyrimidine*-2-$^{14}$C]sulfometuron methyl added to each capsule was 24.5 mg, 49.0 μCi (2 μCi/mg).

A solution of [*phenyl*(U)-$^{14}$C]sulfometuron methyl was prepared as described above. The total [*phenyl*(U)-$^{14}$C]sulfometuron methyl in each of 15 capsules was 22.6 mg, 45 μCi (2 μCi/mg).

**Dosing.** Two 40-kg goats were acclimated for 8–12 days prior to dosing. Following the acclimation period, a balling gun was used to administer the capsule (containing 23–25 mg of [$^{14}$C]-sulfometuron methyl) twice daily (after the morning and evening milkings) for 7 consecutive days. Due to the difference in the daily consumption of each goat, the dietary dosage levels were 25 ppm for the [*pyrimidine*-2-$^{14}$C]sulfometuron methyl dosed goat and 60 ppm for the [*phenyl*(U)-$^{14}$C]sulfometuron methyl dosed goat. The [$^{14}$C]sulfometuron methyl in each spare capsule was removed, dissolved in mobile phase, and analyzed by high-performance liquid chromatography (HPLC) to determine radiopurity (gradient 1).

**Collection of Samples.** Milk was collected twice daily and frozen; urine and feces were collected once daily and frozen (volumes/weights were recorded). The goats were sacrificed approximately 20 h after the last dose. Organs and tissues were harvested including blood, heart, lungs, liver, kidneys, pancreas, muscle (loin, flank, and leg), fat (renal, omental, peripheral, and back), and intestinal and stomach contents.

**Determination of Total $^{14}$C-Labeled Residues.** Aliquots (1 mL) of each milk and urine sample were analyzed directly by LSC. Feces, organs, and tissues were lyophilized and homogenized in a Waring Blendor (Dynamics Corp. of America, New Hartford, CT). Aliquots (0.2–0.5 g each) were combusted using a Packard Model 306 sample oxidizer (Packard Instrument Co., Downers Grove, IL) and analyzed by LSC to determine the total $^{14}$C-labeled residues in each sample.

**Extraction and Characterization of $^{14}$C-Labeled Residues from Milk, Excreta, Organs, and Tissues.** *High-Performance Liquid Chromatography Methods.* Aliquots of extracts were analyzed using a Hewlett-Packard Model 1090 liquid chromatograph (Hewlett-Packard Co., Avondale, PA) with a Du Pont Zor-

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**Table I. HPLC Retention Times (RT) for Sulfometuron Methyl and Metabolites[a]**

| gradient 1[b] | | gradient 2[b] | |
|---|---|---|---|
| compound[c] | RT, min | compound[c] | RT, min |
| sulfometuron methyl | 20.2 | sulfometuron methyl | 26.1 |
| HM-PY-SM[d] | 17.9 | HM-PY-SM | 23.8 |
| sulfonamide | 15.8 | pyrimidinyl urea | 21.2 |
| FA-sulfonamide | 14.7 | pyrimidinamine | 13.2 |
| saccharin | 8.4 | HM-PA[e] | 10.3 |

[a] Structures are shown in Figure 3. [b] HPLC conditions and gradients are described in the text. [c] The chemical names for the compounds are as follows: sulfometuron methyl, methyl 2-[[[[(4,6-dimethyl-2-pyrimidinyl)amino]carbonyl]amino]sulfonyl]benzoate; HM-PY-SM, 2-methyl 2-[[[[(4-(hydroxymethyl)-6-methyl-2-pyrimidinyl]aminocarbonyl]amino]sulfonyl]benzoate; sulfonamide, methyl 2-(aminosulfonyl)benzoate; FA-sulfonamide, 2-(aminosulfonyl)-benzoic acid; saccharin, 1,2-benzisothiozol-3(2*H*)-one 1,1-dioxide; pyrimidine urea, *N*-(4,6-dimethyl-2-pyrimidinyl)urea; pyrimidine amine, (4,6-dimethyl-2-pyrimidinyl)amine; HM-PA, 2-amino-6-methyl-4-pyrimidine methanol. [d] HM-PY-SM, (hydroxymethyl)pyrimidine sulfometuron methyl. [e] HM-PA, (hydroxymethyl)pyrimidinamine.

bax C-18 analytical column at 40 °C and a flow rate of 1 mL/min. One of the following two gradients was used depending on the position of the radiolabel:

| time, min | % $CH_3CN$ | % pH 2.2 $H_2O$ (acetic acid) |
|---|---|---|
| | Gradient 1 | |
| 0 | 10 | 90 |
| 8 | 10 | 90 |
| 20 | 65 | 35 |
| 25 | 95 | 5 |
| 28 | 95 | 5 |
| 32 | 10 | 90 |
| | Gradient 2 | |
| 0 | 0 | 100 |
| 5 | 0 | 100 |
| 6 | 10 | 90 |
| 14 | 10 | 90 |
| 26 | 65 | 35 |
| 31 | 95 | 5 |
| 34 | 95 | 5 |
| 38 | 0 | 100 |

Gradients 1 and 2 were used for *phenyl*(U)-$^{14}$C- and *pyrimidine*-2-$^{14}$C-labeled samples, respectively.

Fractions were collected at 0.5-min intervals and analyzed by LSC using a Mark III liquid scintillation counter, Model 6881 (Tracor Analytical, Inc., Elk Grove, IL). Metabolites were identified by cochromatography of unlabeled standards. Retention times for sulfometuron methyl and metabolites are shown in Table I.

*Stability.* At room temperature, sulfometuron methyl is hydrolytically stable at pH 5 (half-life of 14 days) and pHs 7 and 9 (half-life of >30 days) (Brattsten, 1987). The stability of sulfometuron methyl was tested at pH 2.5 and was not found to degrade after several hours at room temperature. Due to its hydrolytic stability, no significant decomposition of sulfometuron methyl should occur during the sample extraction procedures described in the following sections. To assure stability, all extracts were stored at −20 °C prior to HPLC analyses.

*Milk.* A flow sheet for extraction and characterization of $^{14}$C-labeled residues in milk is shown in Figure 1. A milk composite from each goat was prepared by combining 60 mL of milk from each of the morning and evening milkings.

*Urine.* A composite urine sample from each goat was prepared by mixing 10 mL of urine from each sample collected on days 1–7. An equal volume of methanol was added to 15 mL of the urine composite to precipitate salts. The mixture was refrigerated overnight and centrifuged at 21 000 rpm, and the solids were discarded. The methanol was removed by rotary evaporation. The aqueous urine was adjusted to pH 2.5 with 1 M HCl and immediately partitioned three times with equal volumes of dichloromethane. The dichloromethane and aqueous phases were concentrated and analyzed by HPLC as previously described.

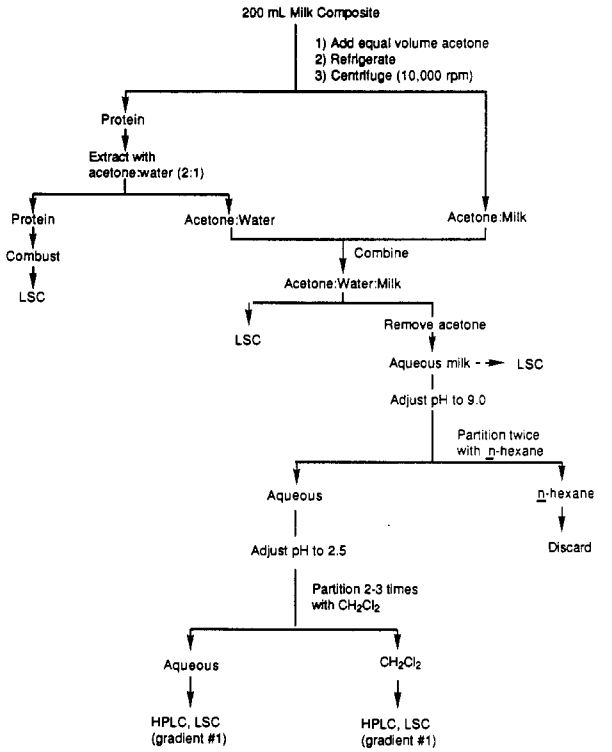


**Figure 1.** Flow sheet depicting extraction and analyses of milk.

*Feces.* A feces composite was prepared by mixing 10–20-g aliquots of lyophilized feces from each sample collected on days 1–7. Twenty grams of lyophilized feces composite from each goat was extracted from two to seven times with *n*-hexane, dichloromethane/methanol (3:1 v/v), and methanol, respectively. Finally the feces were continuously extracted with methanol using a Soxhlet apparatus. The remaining pulp was combusted to determine the amount of unextractable $^{14}$C-labeled residues.

The $^{14}$C-labeled residues in the extracts were purified and concentrated by rotary evaporation followed by nitrogen evaporation. Each purified extract was analyzed by HPLC as previously described.

*Liver, Muscle, and Kidney.* A flow sheet for analyses of liver, muscle, and kidney is shown in Figure 2. A muscle composite from each goat was prepared by mixing approximately 15 g each of lyophilized loin, leg, and flank muscle samples. Liver, muscle composite, or kidney was extracted and analyzed as shown on the flow sheet in Figure 2.

Details of the proteolytic enzyme digestion of extracted tissue follow. Following the buffer and acetone/buffer extractions of 40–90 g of fresh tissue, the tissue was treated with proteolytic enzyme to release any $^{14}$C-labeled residues that might be associated with proteins. The extracted tissue was lyophilized and rinsed with pH 7.4 buffer to remove any remaining solvents that might destroy the protease. The tissue was hydrated with 50–100 mL of pH 7.4 buffer and divided into two portions. Protease (100–200 mg of type XIV from *Streptomyces griseus*, Sigma Chemical Co., St. Louis, MO) was dissolved in 5 mL of pH 7.4 buffer and added to one portion of tissue; the portion containing no enzyme served as the control. The tissue samples were incubated overnight at 37 °C. Each sample was centrifuged at 10 000 rpm for 15–20 min at 3 °C, and the supernatant was decanted and analyzed by LSC.

Gradients 1 and 2 were used for HPLC analysis of extracts and protease-digested material.

**Stability of [$^{14}$C]Sulfometuron Methyl and [$^{14}$C](Hydroxymethyl)pyrimidine Sulfometuron Methyl in Proteolytic Enzyme.** [$^{14}$C]Sulfometuron methyl and [$^{14}$C](hydroxymethyl)pyrimidine sulfometuron methyl did not undergo bridge cleavage upon exposure to proteolytic enzyme. It appears that protease is not able to attack the sulfonylurea bridge. This indicates that the protein-bound $^{14}$C metabolites in liver were

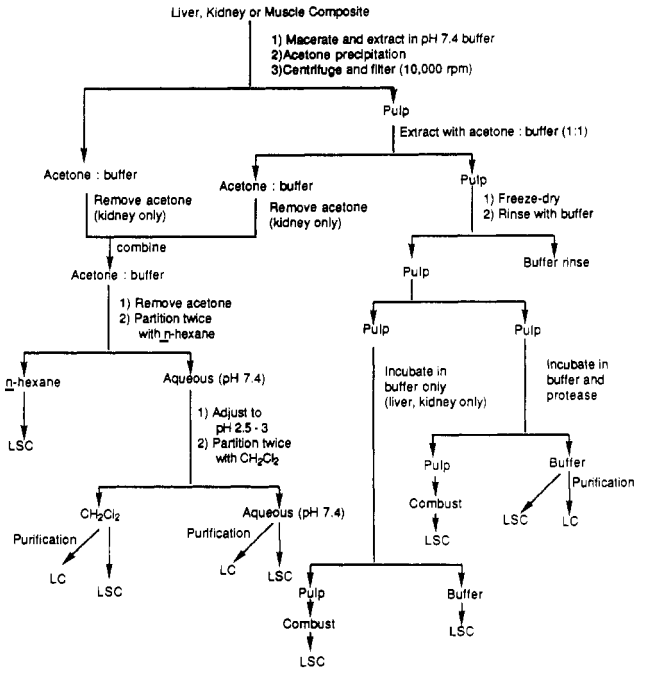


**Figure 2.** Flow sheet depicting extraction and analyses of liver, kidney, or muscle.

probably not primarily parent or (hydroxymethyl)pyrimidine sulfometuron methyl because each one is stable under protease digestion.

**Method To Determine If $^{14}$C-Labeled Residues Released by Protease in Liver Are Associated with Proteins.** A modified method, described by W. C. Sneiders, that separates lipids, salts, nucleic acids, and proteins in animal tissues was used to determine if unextractable $^{14}$C-labeled residues could be released by proteolytic enzyme.

Liver was extracted twice with dichloromethane/methanol (2:1 v/v) to remove lipids from the tissue. The liver was then extracted twice with cold trichloroacetic acid (TCA) to remove salts and soluble cofactors from the tissue. Nucleic acids were removed by heating the liver for 15 min in 90 °C TCA. The liver pellet was rinsed with TCA. Total $^{14}$C-labeled residues in each of these extracts were determined by LSC analysis. The remaining liver pellet consisted of only proteins. The liver pellet was combusted to determine the amounts of $^{14}$C-labeled residues associated with proteins in the liver.

**Fat.** Lyophilized, peripheral fat tissue (from the [*pyrimidine*-2-$^{14}$C]sulfometuron methyl dosed goat) was macerated in *n*-hexane using a Tekmar Tissumizer (Tekmar Co., Cincinnati, OH) and extracted by shaking for 1 h. The fat/hexane mixture was centrifuged and filtered, and the pellet was extracted three more times with *n*-hexane. Next the fat was extracted with pH 7.4 phosphate buffer followed by buffer/acetone (1:1 v/v). These two extracts were combined, and the acetone was removed by rotary evaporation. The combined extract was partitioned with *n*-hexane and then dichloromethane to remove oils. The aqueous phase was adjusted to pH 2.5 and partitioned with dichloromethane; the dichloromethane was analyzed by HPLC as previously described.

## RESULTS AND DISCUSSION

**Distribution of $^{14}$C-Labeled Residues.** The majority of dosed radioactivity, 94–99%, was excreted in urine and feces (Table II). Less than 1% of dose was excreted in the milk. Levels of $^{14}$C-labeled residues in milk and edible tissues are shown in Table III. Levels of $^{14}$C-labeled residues in the milk, muscle, and fat were low. As expected, the liver and kidney contained the highest levels of $^{14}$C-labeled residues on a fresh weight basis—0.35–0.72 and 0.14–0.21 ppm, respectively.

**Composition of $^{14}$C-Labeled Residues in Urine and Feces.** The majority of $^{14}$C-labeled residues in urine consisted of (hydroxymethyl)pyrimidine sulfometuron methyl (56–60% of the dosed radioactivity), indicating rapid hydroxylation of sulfometuron methyl (Tables IV and V). Intact sulfometuron methyl accounted for approximately 10% of the dosed radioactivity. ([$^{14}$C]Sulfometuron methyl was stable in the dosing capsules as evidenced by 97–99% radiopurity upon analysis of the spare capsule contents following dosing.) Each minor metabolite found in either the [*phenyl*(U)-$^{14}$C]- or [*pyrimidine*-2-$^{14}$C]sulfometuron methyl dosed goat, including sulfoamide, saccharin, and (hydroxymethyl)pyrimidinamine, accounted for <8% of the dosed radioactivity. These metabolites are probably a result of bridge cleavage of (hydroxymethyl)pyrimidine sulfometuron methyl.

Sulfometuron methyl and (hydroxymethyl)pyrimidine

**Table II. Recovery of Excreted $^{14}$C-Labeled Residues from Goats Dosed with [$^{14}$C]Sulfometuron Methyl**

| | % of total dose excreted | |
|---|---|---|
| sample | [$^{14}$C]phenyl | [$^{14}$C]pyrimidine |
| milk | <1 | <1 |
| urine | 83 | 84 |
| feces | 11 | 15 |
| total | 94 | 99 |

**Table III. $^{14}$C-Labeled Residue Levels in Edible Tissues and Organs at Sacrifice of Goats**

| | total ppm | |
|---|---|---|
| sample | [$^{14}$C]phenyl[a] | [$^{14}$C]pyrimidine[b] |
| milk | 0.06 | 0.06 |
| kidney | 0.14 | 0.21 |
| liver | 0.72 | 0.35 |
| fat | ND[c] | 0.04 |
| muscle | 0.04 | 0.03 |

[a]Dosed at 60 ppm in diet. [b]Dosed at 25 ppm in diet. [c]ND, not detectable, ≤0.005 ppm.

**Table IV. Composition of $^{14}$C-Labeled Residues in Urine and Feces from Goat Dosed with [*phenyl*(U)-$^{14}$C]-Sulfometuron Methyl**

| | % of total dose | |
|---|---|---|
| compound[a] | urine | feces |
| sulfometuron methyl (SM) | 10 | 2 |
| HM-PY-SM | 60 | 4 |
| sulfonamide | 7 | 1 |
| FA-sulfonamide | ND[b] | <1 |
| saccharin | 2 | ND |
| unknowns[c] | 4 | 1 |
| bound | NA[d] | 2 |
| total | 83 | 11 |

[a] Structures are shown in Figure 3. [b] ND, not detected. [c] Unknowns consist of two or more $^{14}$C metabolites whose identity could not be confirmed. [d] NA, not applicable.

**Table V. Composition of $^{14}$C-Labeled Residues in Urine and Feces from Goat Dosed with [*pyrimidine*-2-$^{14}$C]-Sulfometuron Methyl**

| | % of total dose | |
|---|---|---|
| compound[a] | urine | feces |
| sulfometuron methyl (SM) | 13 | 4 |
| HM-PY-SM | 57 | 4 |
| (hydroxymethyl)pyrimidinamine | 5 | <1 |
| unknowns[b] | 9 | 3 |
| bound | NA[c] | 3 |
| total | 84 | 15 |

[a] Structures are shown in Figure 3. [b] Unknowns consist of two or more $^{14}$C metabolites whose identity could not be confirmed. [c] NA, not applicable.

Table VI. Extraction of $^{14}C$-Labeled Residues from Milk

| extraction phases | % of total $^{14}C$-labeled residues[a] | |
|---|---|---|
| | [$^{14}C$]phenyl | [$^{14}C$]pyrimidine |
| dichloromethane | 66 | 62 |
| aqueous | 28 | 34 |
| protein (bound) | 6 | 4 |
| total | 100 | 100 |

[a] Values for $^{14}C$-labeled residues were normalized; the average recovery of radioactivity from the initial sample to the analyzed fraction was 98%.

Table VII. Distribution of $^{14}C$-Labeled Residues in Extracts of Liver, Kidney, Muscle, and Fat

| | extraction phases | | | |
|---|---|---|---|---|
| | buffer[a] | protease solubilized[a] | bound | total |
| liver | | | | |
| [*phenyl*-$^{14}C$]SM[b] | 24 | 70 | 6 | 100 |
| [*pyrimidine*-$^{14}C$]SM[c] | 44 | 50 | 6 | 100 |
| kidney | | | | |
| [*phenyl*-$^{14}C$]SM | 59 | 39 | 2 | 100 |
| [*pyrimidine*-$^{14}C$]SM | 70 | 25 | 5 | 100 |
| muscle | | | | |
| [*phenyl*-$^{14}C$]SM | 64 | 26 | 10 | 100 |
| [*phenyl*-$^{14}C$]SM | 53 | 38 | 9 | 100 |
| fat[d] | 93 | NU[e] | 7 | 100 |

[a] Values for $^{14}C$-labeled residues were normalized; the recoveries of radioactivity from the initial sample to the analyzed fraction ranged from 92 to 100%. [b] Tissue/organ from goat dosed with [*phenyl*(U)-$^{14}C$]sulfometuron methyl. [c] Tissue/organ from goat dosed with [*pyrimidine*-2-$^{14}C$]sulfometuron methyl. [d] Peripheral fat from [*pyrimidine*-2-$^{14}C$]sulfometuron methyl dosed goat. [e] NU, not used.

sulfometuron methyl were the major components (2–4% of the dosed radioactivity) in the feces (Tables IV and V). As in the urine, the feces contained small amounts of sulfonamide, saccharin, and (hydroxymethyl)pyrimidinamine, depending on the $^{14}C$-labeled position.

**Extraction and Composition of $^{14}C$-Labeled Residues in Milk.** The majority of the $^{14}C$-labeled residues in the milk (62–66%) were relatively nonpolar and partitioned from the aqueous milk into dichloromethane (Table VI). Only 4–6% of the $^{14}C$-labeled residues were associated with the milk protein. Sulfometuron methyl consisted of <10% (<0.01 ppm) of the $^{14}C$-labeled residues in the milk. HPLC results of the aqueous or dichloromethane fraction showed that (hydroxymethyl)pyrimidine sulfometuron methyl was the major metabolite in milk, amounting to 54–57% (0.03 ppm) of the total $^{14}C$-labeled residues in milk. Other minor metabolites included sulfonamide, saccharin, and (hydroxymethyl)pyrimidinamine. These are probably a result of the sulfonylurea bridge cleavage of (hydroxymethyl)pyrimidine sulfometuron methyl.

**Extraction and Composition of $^{14}C$-Labeled Residues in Edible Organs/Tissues.** *Liver.* Only 24–44% of the $^{14}C$-labeled residues in the liver were extractable with pH 7.4 buffer (Table VII). The remaining unextractable $^{14}C$-labeled residues were released upon incubation of the buffer-extracted liver with proteolytic enzyme. Significant $^{14}C$-labeled residues were not released without the proteolytic enzyme, indicating that protease was necessary to release unextractable $^{14}C$-labeled residues. As shown in Table VIII, 45% of the 1$^{14}C$-labeled residues in the [*pyrimidine*-2-$^{14}C$]sulfometuron methyl liver and 72% of the $^{14}C$-labeled residues in the [*phenyl*(U)-$^{14}C$]-sulfometuron methyl liver were associated with proteins. This corresponds very well with the amounts of $^{14}C$-labeled residues that were released from the liver upon protease

Table VIII. Amount of $^{14}C$-Labeled Residues in Liver Associated with Protein, Nucleic Acids, Salts, and Lipids

| extraction phases | tissue fraction | % of total $^{14}C$-labeled residues | |
|---|---|---|---|
| | | phenyl[a] | pyrimidine[b] |
| $CH_2Cl_2$:$CH_3OH$ | lipids | 13 | 40 |
| cold TCA[c] | salts[d] | 3 | 6 |
| hot TCA | nucleic acids | 10 | 7 |
| TCA rinse | nucleic acids | 2 | 2 |
| nonextractable | proteins | 72 | 45 |
| total | | 100 | 100 |

[a] Liver from [*phenyl*(U)-$^{14}C$]sulfometuron methyl dosed goat. [b] Liver from [*pyrimidine*-2-$^{14}C$]sulfometuron methyl dosed goat. [c] TCA, trichloroacetic acid. [d] Includes salts and soluble cofactors.

Table IX. Composition of $^{14}C$-Labeled Residues in Liver, Kidney, and Muscle from Goat Dosed with [*phenyl*(U)-$^{14}C$]-Sulfometuron Methyl

| compound[a] | % of total $^{14}C$-labeled residues | | |
|---|---|---|---|
| | liver | kidney | muscle |
| sulfometuron methyl (SM) | 9 | 10 | 20 |
| HM-PY-SM | 5 | 34 | 25 |
| sulfonamide | 2 | 17 | ND[b] |
| saccharin | 37 | 28 | ND |
| FA-sulfonamide | 30 | ND | ND |
| unknowns[c] | 11 | 9 | 45 |
| bound | 6 | 2 | 10 |
| total | 100 | 100 | 100 |

[a] Structures are shown in Figure 3. [b] ND, not detected. [c] Unknowns consist of two or more $^{14}C$ metabolites whose identity could not be confirmed.

Table X. Composition of $^{14}C$-Labeled Residues in Liver, Kidney, Muscle, and Fat from Goat Dosed with [*pyrimidine*-2-$^{14}C$]Sulfometuron Methyl

| compound[a] | % of total $^{14}C$-labeled residues | | | |
|---|---|---|---|---|
| | liver | kidney | muscle | fat |
| sulfometuron methyl (SM) | 9 | 24 | 17 | 18 |
| HM-PY-SM | 6 | 45 | 17 | 18 |
| pyrimidinylurea | 10 | 3 | ND[b] | ND |
| pyrimidinamine | 20 | 5 | ND | ND |
| (hydroxymethyl)pyrimidine amine | 18 | 8 | ND | ND |
| unknowns[c] | 31 | 10 | 57 | 57 |
| bound | 6 | 5 | 9 | 7 |
| total | 100 | 100 | 100 | 100 |

[a] Structures are shown in Figure 3. [b] ND, not detectable, ≤0.005 ppm. [c] Unknowns consist of two or more $^{14}C$ metabolites whose identity could not be confirmed.

treatment [50 and 70% [*pyrimidine*]- and [*phenyl*]sulfometuron methyl, respectively (Table VII)]. This indicates that the protease-solubilized $^{14}C$-labeled residues were probably associated with proteins in the liver and not nucleic acids.

Sulfometuron methyl consisted of <10% (0.03–0.07 ppm) of the $^{14}C$-labeled residues in the liver (Tables IX and X). Unlike the milk and urine, only small amounts of (hydroxymethyl)pyrimidine sulfometuron methyl (5–6% of total $^{14}C$-labeled residues) were detected in the liver. The major components consisted of hydrolytic "fragments" of the intact parent or (hydroxymethyl)pyrimidine sulfometuron methyl including saccharin, FA-sulfonamide, pyrimidinylurea, pyrimidinamine, and (hydroxymethyl)-pyrimidinamine, which were recovered after protease enzyme treatment of the liver.

*Kidney.* About 59–70% of the $^{14}C$-labeled residues in the kidney were extractable with pH 7.4 buffer (Table VII). The remaining unextractable $^{14}C$-labeled residues were released with proteolytic enzyme upon incubation of

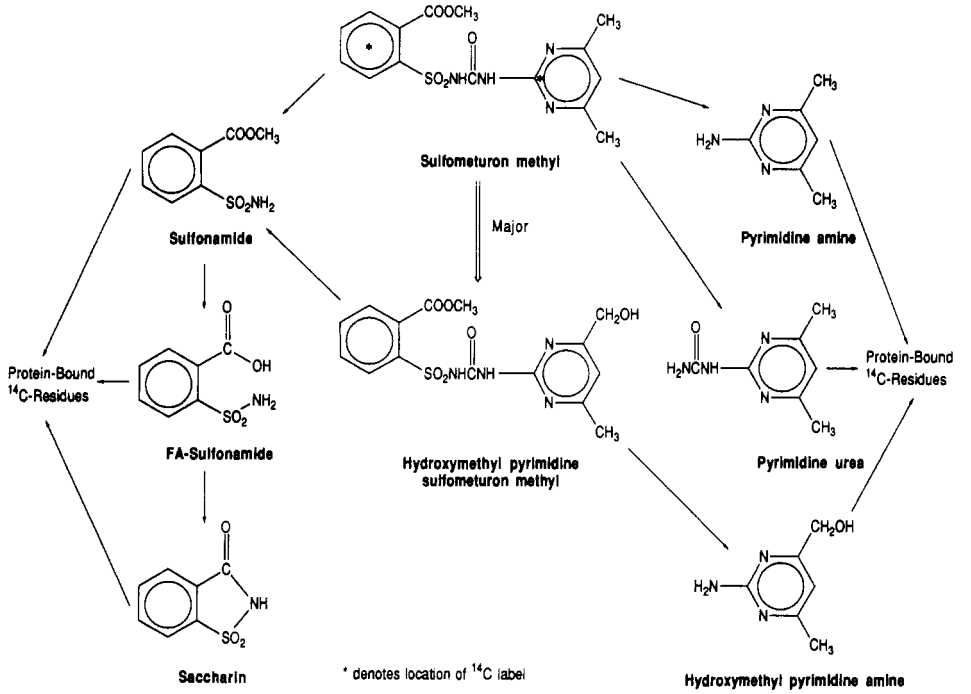


**Figure 3.** Proposed metabolic pathway of sulfometuron methyl in the goat.

the buffer-extracted kidney. Significant $^{14}$C-labeled residues were not released without proteolytic enzyme, indicating that protease was necessary to release these bound $^{14}$C-labeled residues.

Sulfometuron methyl consisted of 10–24% (0.01–0.05 ppm) of the $^{14}$C-labeled residues in the kidney (Tables IX and X). As in the milk and urine, (hydroxymethyl)pyrimidine sulfometuron methyl is the major metabolite in the kidney, amounting to 34–45% (0.04–0.09 ppm) of the total $^{14}$C-labeled residues in the kidney. The majority of the saccharin, pyrimidinylurea, pyrimidinamine, and (hydroxymethyl)pyrimidinamine was detected in the protease extract.

*Muscle.* About 53–64% of the $^{14}$C-labeled residues in the muscle was extractable with pH 7.4 buffer (Table VII). The remaining unextractable $^{14}$C-labeled residues were released with proteolytic enzyme upon incubation of the buffer-extracted muscle. Sulfometuron methyl consisted of 17–20% (<0.01 ppm) of the $^{14}$C-labeled residues in the muscle (Tables IX and X). Although sulfometuron methyl was present in muscle, accumulation was insignificant. A similar amount of (hydroxymethyl)pyrimidine sulfometuron methyl was also detected in the muscle.

*Fat.* All fat tissues contained insignificant amounts of $^{14}$C-labeled residues (Table III), except the peripheral fat from the [*pyrimidine*-2-$^{14}$C]sulfometuron methyl dosed goat. The peripheral fat was extracted, and $^{14}$C-labeled residues in some extracts were characterized.

More than 90% of the $^{14}$C-labeled residues were extractable with pH 7.4 buffer (Table VII). Most of these extractable $^{14}$C-labeled residues were relatively nonpolar and partitioned from the aqueous buffer into *n*-hexane or dichloromethane. Total $^{14}$C-labeled residue levels in the peripheral fat were relatively low, amounting to <0.04 ppm.

Sulfometuron methyl consisted of 18% (<0.01 ppm) of the $^{14}$C-labeled residues in the peripheral fat (Table X). A similar amount of (hydroxymethyl)pyrimidine sulfometuron methyl was also detected. Nonpolar $^{14}$C-labeled residues (43%, 0.02 ppm) present in the *n*-hexane and dichloromethane extracts could not be analyzed by HPLC due to excessive amounts of oils.

**Proposed Metabolic Pathway.** The major metabolic pathway of sulfometuron methyl in goats is via rapid hydroxylation of the methylpyrimidine portion of the molecule, producing (hydroxymethyl)pyrimidine sulfometuron methyl (Figure 3). (Hydroxymethyl)pyrimidine sulfometuron methyl then undergoes cleavage of the sulfonylurea bridge, producing the sulfonamide and (hydroxymethyl)pyrimidinamine. In edible tissues, significant amounts of $^{14}$C-labeled compounds become protein bound, particularly in the liver. These protein bound (or conjugated) $^{14}$C metabolites could not be characterized without destruction of the $^{14}$C metabolite.

**Conclusions.** The metabolic fate of sulfometuron methyl was examined in lactating goats using sulfometuron methyl labeled separately with $^{14}$C at the phenyl and pyrimidine moieties. The dosed sulfometuron methyl was rapidly metabolized to (hydroxymethyl)pyrimidine sulfometuron methyl and eliminated via urinary and fecal excreta. Approximately 94–99% of dosed radioactivity was recovered in the excreta. The maximum concentration of $^{14}$C-labeled residues excreted in the milk was approximately 0.07 ppm on the fourth dosing day. Less than 10% (<0.01 ppm) of the total $^{14}$C-labeled residues in milk was parent, which indicates that accumulation of sulfometuron methyl in milk is minimal. Accumulation of $^{14}$C-labeled residues in meat and fat was also negligible (≤0.03 ppm). As expected, the liver and kidney did contain significant $^{14}$C-labeled residues, 0.35–0.72 and 0.14–0.21 ppm, respectively.

The majority of the $^{14}$C-labeled residues in the liver and kidney were protein-bound and could only be released after proteolytic enzyme treatment. Parent and (hydroxymethyl)pyrimidine sulfometuron methyl were identified as the major buffer-extractable $^{14}$C-labeled residues. Saccharin, pyrimidinylurea, pyrimidinamine, and (hydroxymethyl)pyrimidinamine were characterized as the major protease-solubilized $^{14}$C-labeled compounds. These $^{14}$C-labeled compounds were apparently associated with the protein in the liver and kidney. Parent and (hydroxymethyl)pyrimidine sulfometuron methyl were stable in the presence of protease; protease apparently does not attack

the sulfonylurea bridge when the sulfonylurea is not "bound" to protein.

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